CALIFORNIA ENVIRONMENTAL PROTECTION AGENCY
DEPARTMENT OF PESTICIDE REGULATION
MEDICAL TOXICOLOGY BRANCH

SUMMARY OF TOXICOLOGY DATA
PROPOXYCARBAZONE-SODIUM

Chemical Code # 5876, Tolerance # 52958
SB 950 # NA.

1 September 2005

I. DATA GAP STATUS

Combined, rat:	No data gap, no adverse effect
Chronic toxicity, dog:	No data gap, no adverse effect
Oncogenicity, mouse:	No data gap, no adverse effect
Reproduction, rat:	No data gap, no adverse effect
Teratology, rat:	No data gap, no adverse effect
Teratology, rabbit:	No data gap, no adverse effect
Gene mutation:	No data gap, no adverse effect
Chromosome effects:	No data gap, no adverse effect
DNA damage:	No data gap, no adverse effect
Neurotoxicity:	Not required at this time

Toxicology one-liners are attached.

All record numbers through 214254 were examined.
** indicates an acceptable study.
Bold face indicates a possible adverse effect.
indicates a study in review.
File name: T050901
Prepared by H. Green, 9/1/05

II. TOXICOLOGY ONE-LINERS AND CONCLUSIONS

These pages contain summaries only. Individual worksheets may contain additional effects.

COMBINED, RAT

**52958-0014, 0044 214195, 214228, "Technical Grade MKH 6561: A Combined Chronic Toxicity/Oncogenicity Testing Study in the Rat", (B.S. Wahle and W.R. Christenson, Bayer Corporation, Agriculture Division, Toxicology, Stilwell, KS., Bayer Report No. 108361, 1 July 1999). 50 (CDF[F-344]/BR) rats per sex per group received MKH 6561 (97.6% propoxycarbazone-sodium) in the diet at 0 (rodent ration with corn oil), 1000, 10000, and 20000 ppm for 24 months. Additional treatment groups of 10 (low and mid-dose) and 20 (control and high dose) per sex per group scheduled for interim sacrifice were necropsied at 12 months. Initial dietary concentrations at the low, middle, and high dose were calculated on a per bodyweight basis (as 50, 500, and 1000 mg/kg/day) for the first 7 months. Thereafter, test diets were prepared at fixed concentrations (nominal ppm). The mean dietary intake of MKH 6561 for 104 weeks was 43 ± 6, 459 ± 37, and 924 ± 68 mg/kg/day for males and 49 ± 2, 525 ± 28, and 1049 ± 54 mg/kg/day for females at the low, mid, and high dose levels respectively. Group mean bodyweight and bodyweight gain were reduced ($p \leq 0.05$) at the mid and high dose levels for interim (through 12 months) and terminal sacrifice animals (24 months). Urine pH was increased in 10000 and 20000 ppm males and females at 3, 6, 12, 18, and 24 months. Decreases in relative liver and adrenal weights were noted for 10000 ppm males and 20000 ppm males and females at terminal sacrifice without gross pathology or significant microscopic changes. Microscopy revealed an increase in mineralization of the renal pelvis in males and females at 20000 ppm at terminal sacrifice (possible correlation to elevated urine pH). Food consumption, clinical observations, ophthalmology, hematology, and serum chemistry were unremarkable. Chronic NOEL = 10000 ppm (459 and 525 mg/kg/day for males and females respectively) based on decreased bodyweight and bodyweight gain at the high dose level. No increase in neoplastic lesions. No adverse effects. Record 214228 in 52958-0044 addressed the toxicological significance of the increase in renal pelvis mineralization at 20000 ppm. Lack of functional impairment and normal aging were cited. Serum-chemistry markers of renal function, blood urea-nitrogen (BUN) and creatinine, at the high dose were generally not increased compared to controls and liver enzymes remained in the normal range. Additionally, it was a common finding for aging F344 rats in the historical control data presented and no incidence differences between control and treated groups were seen in the 1 year sacrifice animals in the present study. A copy of the article "Renal Mineralization- A Ubiquitous Lesion in Chronic Rat Studies" (G.H. Lord and P.M. Newbernet, Mallory Institute of Pathology of Boston, Boston, MA., in Fd Chem. Toxic., Vol. 28, No. 6, pp. 449-455, 1990) was included. No toxicological significance was indicated. No change to the main study status. Acceptable. (Green and Gee, 8/1/05).

**52958-0050 214234, "MKH 6561, Study for Subchronic Oral Toxicity in Rats (Feeding Study for 14 Weeks with a 4-Week Recovery Period)", (W. Bomann, P. Andrews, and B. Schilde, Bayer AG, Department of Toxicology, Wuppertal, Germany, Bayer AG Report No. 25597, Bayer Report No. 108881, 26 September 1996). 10 Wistar rats per sex per group received MKH 6561(97.80% propoxycarbazone) in the diet at 0 (standard diet), 250, 1000, 4000, and 20000 ppm for 14 weeks. Ten additional control and high dose rats per sex per group received test diets for 14 weeks followed by a 4-week recovery phase where they received untreated standard diet. The mean dietary intake of MKH 6561 was 17.4, 73.0, 286.4, and 1507.5 mg/kg/day for males and 21.6, 81.6, 350.6, and 1769.9 mg/kg/day for females at 250, 1000, 4000, and 20000 ppm respectively. Water intake was increased (statistically significant) at 20000 ppm for both sexes during treatment and was comparable to controls during the recovery period. Glucose and triglyceride levels were significantly reduced in high dose females. Clinical signs, mortality, bodyweight, food consumption, ophthalmology, hematology, urinalysis, necropsy, and organ weights were not affected by treatment. Microscopy indicated increased incidence of vacuoles in

the squamous epithelium of the forestomach along with minimal epithelial hyperplasia and subepithelial inflammatory infiltration for main group animals that received 20000 ppm. These effects reversed during the 4-week recovery period. NOEL = 4000 ppm (increased water intake, reduced blood triglycerides and glucose in females, and forestomach epithelium irritation). No adverse effects. Acceptable. (Green and Gee, 7/28/05).

CHRONIC TOXICITY, DOG

**52958-0013 214194, "Technical Grade MKH 6561: A Chronic Toxicity Feeding Study in the Beagle Dog", (R.D. Jones and T.F. Hastings, Bayer Corporation (formerly Miles, Inc.), Agriculture Division, Toxicology, Stilwell, KS., Agricultural Division Report No. 108189, 22 September 1998 and D.L. Van Goethem, Report No. 108189-1, 15 January 2001). 4 Beagle dogs per sex per group received MKH 6561 (97.9% propoxycarbazone-sodium) in the diet at 0 (canine diet), 2000, 10000, and 25000 ppm for 12 months. Group average daily consumption of MKH 6561 during the treatment period was 52.2, 258.0, and 630.7 mg x kg^{-1} x day^{-1} for males and 55.7, 235.3, and 605.4 mg x kg^{-1} x day^{-1} for females at 2000, 10000, and 25000 ppm respectively. Food consumption was generally decreased for females at 10000 and 25000 ppm throughout the treatment period. Male food consumption was comparable to control values. Group mean bodyweight was increased for high dose males and for mid-dose females. Absolute male adrenal gland weights were significantly increased at 25000 ppm. Relative adrenal weights were comparable to controls. Female absolute and relative adrenal weights were generally comparable to control values. Relative ($p \leq 0.05$) and absolute (ns) heart weights were reduced at 10000 and 25000 ppm for females. Male absolute heart weights were increased (ns) at the high dose, relative weights were comparable to controls. Heart microscopy was unremarkable. No treatment-related effects were recorded for clinical signs, mortality, ophthalmology, electrocardiography, clinical neurology, serum chemistry, hematology, urinalysis, gross pathology, and histopathology. Chronic NOEL = 2000 ppm (reduced food consumption and reduced relative heart weights in females at the mid and high dose levels). NOAEL = HDT or 25000 ppm (M = 630.7 mg/kg/day, F = 605.4 mg/kg/day). No adverse effects were indicated. Acceptable. (Green and Gee, 7/28/05).

52958-0011 214192, "Technical Grade MKH 6561, A Range-Finding Toxicity Feeding Study in the Beagle Dog", (R. D. Jones and B. F. Hamilton, Bayer Corporation, Agricultural Division, Toxicology, Stilwell, KS., Agricultural Division Report No. 107479, 3 June 1997). 2 Beagle dogs per sex per group received MKH 6561 in the diet at 0 (canine diet), 1000, 5000, 10000, and 40000 ppm for 2 months. Group mean calculated MKH 6561 intake during the treatment period was 27.7, 140.3, 322.2, and 1406.7 mg/kg/day for males and 32.4, 134.1, 285.6, and 1181.2 mg/kg/day for females at 1000, 5000, 10000, and 40000 ppm respectively. One high dose female was euthanized on day 16 in a moribund condition (due to ongoing illness, not treatment). Food consumption and bodyweight were generally reduced at the high dose level in both sexes throughout the treatment period. Poor palatability was indicated in the first few days of treatment. Treatment-related elevation of cytochrome P-450 and N-demethylase activities were noted for males and females respectively at 40000 ppm. Clinical signs, mortality, hematology, urinalysis, gross pathology, organ weights, and histopathology were unremarkable. NOEL = 10000 ppm (decreased food consumption, reduced bodyweight, and elevated liver enzyme activity at 40000 ppm). Unacceptable, not upgradeable (too few animals per group, exposure period of 60 rather 90 days, incomplete histopathology). Supplemental data. (Green and Gee, 7/28/05).

ONCOGENICITY, MOUSE

**52958-0054 214239, "MKH 6561, Oncogenicity Study in $B6C3F_1$ Mice , Dietary Administration Over 2 Years", (L. Schadt, Bayer AG, Department of Toxicology, Wuppertal, Germany, Bayer AG Report No. 28845, Bayer AG Study No. T7060911, Test No. 109266, 14 June 1999). 50 $B6C3F_1$

mice per sex per group received MKH 6561 (97.6% propoxycarbazone-sodium) in the diet at 0 (standard diet), 280, 1400, and 7000 ppm for 107 weeks. Mean MKH 6561 intake during the treatment period was 74.6, 369.0, and 1880.9 mg/kg/day for males and 126.2, 626.9, and 3106.1 mg/kg/day for females at 280, 1400, and 7000 ppm respectively. Male bodyweight was significantly lower compared to controls at 280 (weeks 1-10 only), 1400, and 7000 ppm. Female bodyweight was decreased at 7000 ppm. Relative kidney weights were reduced relative to controls at 1400 ppm ($p \leq 0.05$) and 7000 ppm (ns). Absolute kidney weights were significantly lower than control values for females at 1400 and 7000 ppm and for high dose males. The incidence of malignant lymphomas was increased ($p \leq 0.05$) relative to controls in 1400 ppm females that died or were killed prior to terminal sacrifice. The neoplasms were found in each of nine females that died near the end of the study (weeks 85 through 103). All were coded by the pathologist as the probable cause of death. The incidence in terminal sacrifice animals (weeks 106 and 107) was comparable to controls. The increase in malignant lymphomas for the 1400 ppm females was flagged as a potential adverse effect under CFR (Code of Federal Regulations) 40, 158.34. However, there were no significant increases in the total number of tumors, the number of benign and malignant tumors, nor the number of animals with tumors for groups across the study. In males, the total number of tumors was actually lowest at 7000 ppm and no increase in malignant lymphomas was seen in 7000 ppm females. Additionally, historical control data (covering the previous 7 year period) from feeding studies for $B6CF3_1$ mice from the National Toxicology Program (NTP) indicate that malignant lymphomas were one of the most common spontaneous neoplasms occurring in untreated female $B6CF3_1$ mice (J.K. Haseman, *et al*, Biostatistics Branch, National Institute of Environmental Health Sciences, Research Triangle Park, NC; in *Toxicologic Pathology*, vol. 26, no. 3, pp 428-441, 1998). These factors, along with the age of the animals, and the extended duration of this mouse study (24 months rather than 18 months as per guidelines), leave the toxicological significance of the increase in these neoplasms inconclusive. Chronic NOEL = 1400 ppm (369.0 mg/kg/day (males) and 626.9 mg/kg/day (females)) based on reduced bodyweight in males at 1400 ppm and above. Acceptable. (Green and Gee, 7/29/05)

52958-0021 214205, “MKH 6561, Subchronic Toxicity Study in B6C3F1-Mice (Administration in the Feed Over 14 Weeks)”, (L. Schadt and V. Geiss, Bayer AG, Department of Toxicology, Wuppertal, Germany, Bayer AG Report No. 25796, Bayer AG Study No. T1055704, Test No. 108877, 6 January 1997). 10 $B6C3F_1$ mice per sex per group received MKH 6561 (97.8% propoxycarbazone-sodium) in the diet at 0 (standard diet), 625, 2500, and 10000 ppm for 14 weeks. The average MKH 6561 intake during the treatment period was 205.1, 860.2, and 3926.2 mg/kg/day for males and 306.9, 1158.8, and 5108.9 mg/kg/day for females at 625, 2500, and 10000 ppm respectively. Group mean food consumption was slightly increased for males at 10000 ppm. Male and female bodyweights were significantly lower than controls at 2500 and 10000 ppm. Cholesterol was significantly decreased for 10000 ppm males relative to concurrent controls, but within the historical control range. Other serum chemistry parameters were comparable to controls. The leucocyte count was significantly lower than controls in treated males and in high dose females. No treatment-related changes were recorded for the other hematology parameters. Group mean relative liver weights were reduced (ns) in males at 2500 and 10000 ppm relative to controls. Gross pathology and histopathology were unremarkable. NOEL = 625 ppm based on reduced bodyweights. No adverse effects were indicated. Unacceptable, not upgradeable (incomplete histopathology, no ophthalmology). (Green and Gee, 7/28/05)

REPRODUCTION, RAT

52958-0029 214213,** “MKH 6561 (c.n.: Procarbazone-Sodium), Two-Generation Study in Wistar Rats”, (R. Eiben, Bayer AG, Department of Toxicology, Wuppteral, Germany, Bayer AG

Report No. 28792, Bayer AG Study No. T1061527, Test No. 109096, 26 May 1999). 30 rats per sex per group received MKH 6561 (97.6% propoxycarbazone-sodium) in the diet at 0 (standard diet), 1000, 4000, and 16000 ppm through 2 generations with 1 litter per generation. Treatment for F0 and F1 parental animals began 10 and 13 weeks prior to mating respectively. Group mean daily intake of MKH 6561 for F0 animals during the premating period was 74.8, 297.1, and 1230.7 mg/kg/day for males and 92.7, 373.5, and 1605.3 mg/kg/day for females at 1000, 4000, and 16000 ppm respectively. In F1 parents, the values were 79.6, 322.9, and 1313.9 mg/kg/day for males and 103.8, 413.5, and 1907.5 mg/kg/day for females respectively. Group mean food consumption was increased for 16000 ppm females during the premating period. Food consumption for all other groups was generally comparable to controls. Bodyweight was comparable to controls across all treated F0 and F1 groups. No treatment-related effects were seen in mating performance, fertility, sperm parameters, duration of pregnancy and survival of F0 and F1 parental animals and in the number, weight, growth, and survival of F1 and F2 pups. Slightly lower absolute and relative group mean epididymis weights (ns) were noted for F0 males at 16000 ppm relative to controls. At 1000 and 4000 ppm, absolute and relative values were higher than controls for F0 and F1 animals. Necropsy revealed a significant treatment-related increase in dilated cecum in 4000 ppm (F0) and 16000 ppm (F0 and F1) females. There was no microscopic correlation. Treatment-related increases in focal vacuolation of the forestomach epithelium were noted for F1 males at 4000 ppm and for F0 and F1 males and females at 16000 ppm. Chronic NOEL = 1000 ppm based on increased frequency of dilated cecum at 4000 and 16000 ppm. Reproductive NOEL = 16000 ppm. No adverse reproductive effects. The toxicological significance of the increased focal vacuolation of the forestomach epithelium remains unresolved. Acceptable. (Green and Gee, 8/1/05).

52958-0045 214229, “MKH 6561, One-Generation Study in Wistar Rats”, (R. Eiben, Bayer AG, Department of Toxicology, Wuppertal, Germany, Bayer AG Report No. 26743, Bayer AG Study No. T3060953, Test No. 108872, 16 October 1997). Ten Wistar ICO: WU (IOPSCpb) rats per sex per group received MKH 6561 (97.6% propoxycarbazone-sodium) in the diet at 0 (standard diet), 1000, 5000, and 20000 ppm through one generation with one litter. Treatment of F0 animals began 4 weeks prior to mating. Culled pups were necropsied on lactation day 4; remaining weanlings on day 21. Mean MKH 6561 intake during the premating period was 60.5, 299.6, and 1229.7 mg/kg/day for males and 69.4, 384.1, and 1542.2 mg/kg/day for females at 1000, 5000, and 20000 ppm respectively. Parental clinical signs, mortality, bodyweight, food consumption, organ weights, and macroscopy were not affected by treatment through 20000 ppm. Insemination, fertility, and gestation indices were comparable to controls. Mean live litter size was reduced (8.87 at 2000 ppm vs 11.75 in control) with fewer males per litter at 20000 ppm compared to controls (38% versus 52%). F1 pup weights were generally comparable to controls. Pup clinical signs, bodyweight, and macroscopy were unremarkable. Estimated NOEL = 5000 ppm. (Green and Gee, 7/29/05). No worksheet.

TERATOLOGY, RAT

**52958-0015 214199, “MKH 6561, Developmental Toxicity Study in Rats After Oral Administration”, (B. Stahl, Bayer AG, Department of Toxicology, Wuppertal, Germany, Bayer AG Report No. 26772, Bayer AG Study No. T8054289, Test No. 108875, 27 October 1997). 28 inseminated female Wistar Hsd Cpb:WU rats per group received MKH 6561 (98.0% propoxycarbazone-sodium) by oral gavage at 0 (0.5% aqueous carboxymethylcellulose), 100, 300, and 1000 mg/kg/day on gestation days 6 through 19. No treatment-related effects on maternal appearance, behavior, mortality, food and water consumption, bodyweight, clinical chemistry, hematology, or excreta were indicated. Intrauterine development; gestation rate; the number, sex, and weight of fetuses; and fetal external, visceral, and skeletal findings were unaffected by treatment. Maternal and Developmental NOEL = 1000 mg/kg/day. No teratogenicity. Acceptable. (Green and Gee, 7/29/05).

TERATOLOGY, RABBIT

**52958-0048 214232, "MKH 6561, Developmental Toxicity Study in Rabbits After Oral Administration", (B. Holzum, Bayer AG, Department of Toxicology, Wuppertal, Germany, Bayer AG Report No. 27466, Bayer AG Study No. T0061724, Test No. 108851, 30 April 1998). 22 mated female Himalayan rabbits (CHBB:HM) per group received MKH 6561 (98.1% propoxycarbazone-sodium) by oral gavage at 0 (0.5% carboxymethylcellulose in demineralized water), 20, 100, 500, and 1000 mg/kg/day on gestation days 6 through 28. One female at 500 mg/kg/day aborted on gestation day 29. 18 dams at 1000 mg/kg/day aborted between days 19 and 28. All dams that aborted showed decreased food and water intakes, diarrhea, transient weight loss, and cold ears (starting between gestation days 11 and 22). At 1000 mg/kg/day, an increased incidence of females with alopecia was noted. 16 females at 1000 mg/kg/day showed an anal prolapse for several days and 1/16 revealed swelling of the vulva for 5 days before abortion. There were no maternal deaths. Group mean maternal food consumption was significantly reduced at 500 and 1000 mg/kg/day. Decreased water consumption and decreased urination showed higher incidences at 500 mg/kg/day and was noted for all animals at 1000 mg/kg/day during treatment. Reductions in group mean maternal serum enzyme activities, protein, thyroxine (T4) and triiodothyronine (T3) and an increase in cholesterol were recorded at 1000 mg/kg/day. Necropsy revealed enlarged cecum in 11 females at 500 mg/kg/day and in all dams at 1000 mg/kg/day. Changes in the cecal content (gaseous contents, fluid) were also noted at 1000 mg/kg/day. Two high dose females had solid contents in the stomach and another three showed light discoloration of the small intestine. The 500 mg/kg/day female that aborted (No. 3252) showed distinct lobulation and light discoloration of the liver. Two 1000 mg/kg/day dams also showed distinct liver lobulation. Absolute and relative maternal liver weights were reduced (ns) at 1000 mg/kg/day relative to controls. Maternal NOEL = 100 mg/kg/day (reduced food consumption, water intake, urine output, and abortion with associated findings at 500 and 1000 mg/kg/day). Developmental NOEL = 500 mg/kg/day (abortion at 1000 mg/kg/day). No teratogenicity, no adverse effects. Acceptable. (Green and Gee, 7/29/05).

GENE MUTATION

**52958-0006 214187, "MKH 6561, *Salmonella*/Microsome Test", (R. Gahlmann, Bayer AG, Department of Toxicology, Wuppertal, Germany, Bayer Report No. 22798, Study No. T 3039245, Test No. 107404, 14 January 1994). *Salmonella typhimurium* strains TA98, TA100, TA1535, and TA1537 were exposed (quadruplicate cultures) to MKH 6561, in the presence and absence of S9 activation, at 0 (DMSO), 1.6, 8, 40, 200, 1000, and 5000 µg/plate for 48 hours. 10%, 30%, and 4% S9 fractions were used in the first, second, and third trials respectively. The third trial was +S9 only. Bacterial toxicity was indicated at concentrations above 8 µg/plate. Positive controls were functional. No increase in the mutation frequency. Acceptable. (Green and Gee, 7/27/05).

**52958-0022 214206, "MKH 6561, Mutagenicity Study for the Detection of Induced Forward Mutations in the V79-HPRT Assay *In Vitro*", (S. Brendler-Schwaab, Bayer AG, Department of Toxicology, Wuppertal, Germany, Bayer AG Report No. 25198, Bayer AG Study No. T 0054010, Test No. 108876, 19 June 1996). Chinese hamster V79 cells (4×10^6 per 250 ml flask in culture medium) were exposed (in duplicate) to MKH 6561, in the presence and absence of S9, at 0 (medium), 0 (DMSO), 500, 1000, 2000, 3000, and 4000 μg/ml for 5 hours. Two trials were performed. The expression period of 7 days was followed by plating in selection medium with 6-thioguanine, 3×10^5 cells per dish, 8 dishes per culture for mutant selection. No cytotoxicity was indicated. No increase in forward mutations. Positive controls (EMS (-S9) and DMBA (+S9)) were functional. Acceptable. (Green and Gee, 7/27/05).

**52958-0016 214200, "MKH 8394, *Salmonella*/Microsome Test, Plate Incorporation and Preincubation Method", (B. Herbold, Bayer AG, Department of Toxicology, Wuppertal, Germany,

Bayer AG Report No. 28377, Bayer AG Study No. T 2059864, Test No.108957, 7 January 1999). *Salmonella typhimurium* LT2 strains TA98, TA100, TA102, TA1535, and TA1537 were exposed (in triplicate) to MKH 8394 (98.6%), in the presence and absence of S9, at 0 (DMSO), 16, 50, 158, 500, 1581, and 5000 µg/plate for 48 hours at 37°C. Two trials were performed. The direct plate incorporation method was used in the first trial. In the second trial, cultures were preincubated for 20 minutes (37°C) prior to plating. Bacterial toxicity (reduced titer) was indicated at doses above 50 µg/plate (+S9) in the first trial. Positive controls were functional. No increase in the mutation frequency. Acceptable. Note: these data for MKH 8394 are supplemental to MKH 6561, (Green and Gee, 7/26/05).

**52958-0046 214230, "KTS 9061, Metabolite of MKH 6561, *Salmonella*/Microsome Test, Plate Incorporation and Preincubation Method", (B. Herbold, Bayer AG, Department of Toxicology, Wuppertal, Germany, Bayer AG Report No. 28488, Bayer AG Study No. T 9059906, Test No. 108953, 15 February 1999). Triplicate cultures of *Salmonella typhimurium* LT2 strains TA98, TA100, TA102, TA1535, and TA1537 were exposed to KTS 9601 (98.9%), in the presence and absence of S9, at 0 (DMSO), 16, 50, 158, 500, 1581, and 5000 µg/plate for 48 hours. Two trials were performed. Direct plate incorporation was used in the initial trial. In the repeat assay, cultures were preincubated for 20 minutes in test tubes with KTS 9601 prior to plating and 48 hour incubation. In trial one, bacterial toxicity (reduced titer) was noted at 500 µg/plate and higher. No bacterial toxicity through 5000 µg/plate in the repeat assay. No increase in the mutation frequency. Positive controls were functional. Acceptable as supplemental data to MKH 6561. (Green and Gee, 7/22/05).

52958-0028 214212, "Methylthio Analogue Free Acid - MKH 6561, *Salmonella*/Microsome Test, Plate Incorporation and Preincubation Method", (B. Herbold, Bayer AG, Department of Toxicology, Wuppertal, Germany, Bayer AG Report No. 28914, Bayer AG Study No. T9059960, Test No. 109252, 29 June 1999). *Salmonella typhimurium* LT2 strains TA98, TA100, TA102, TA1535, and TA1537 were exposed (in triplicate) to methylthio analogue free acid - MKH 6561 (98.4%), in the presence and absence of S9 activation, at 0 (dimethylformamide), 0.25, 0.50, 1, 2, 4, and 8 µg/plate for 48 hours in two trials. Direct plate incorporation was used in the first trial. In the repeat trial, cultures were preincubated for 20 minutes prior to plating. Positive controls were functional. No increase in the mutation frequency. Acceptable as supplemental data to MKH 6561. (Green and Gee, 7/25/05).

52958-0033 214217, "KTS 9304, *Salmonella*/Microsome Test, Plate Incorporation and Preincubation Method (MKH 6561 - Metabolite)". (B. Herbold, Bayer AG, Department of Toxicology, Wuppertal, Germany, Bayer AG Report No. 28413, Bayer AG Study No. T 7059869, Test No. 108892, 7 January 1999). Triplicate cultures of *Salmonella typhimurium* LT2 strains TA98, TA100, TA102, TA1535, and TA1537 were exposed to KTS 9304, in the presence and absence of S9, at 0 (DMSO), 16, 50, 158, 500, 1581, and 5000 µg/plate for 48 hours at 37°C. Two trials were performed. Direct plate incorporation was used in the first trial. In the second trial, cultures were preincubated for 20 minutes (37°C) prior to plating. Positive controls were functional. No increase in the mutation frequency. Acceptable as supplemental data to MKH 6561. (Green and Gee, 7/26/05).

52958-0032 214216, "4-OH-Saccharine, Soil Metabolite of MKH 6561, *Salmonella*/Microsome Test, Plate Incorporation and Preincubation Method", (B. Herbold, Bayer AG, Department of Toxicology, Wuppertal, Germany, Bayer AG Report No. PH-28646, Bayer AG Study No. T 8059941, Test No. 109076, 13 April 1999). Triplicate cultures of *Salmonella typhimurium* LT2 strains TA98, TA100, TA102, TA1535, and TA1537 were exposed to 4-OH-Saccharine (97.7%), in the presence and absence of S9, at 0 (DMSO), 16, 50, 158, 500, 1581, and 5000 µg/plate for 48 hours at 37°C. Two trials were performed. Direct plate incorporation was used in the first trial. In the second trial, cultures were preincubated for 20 minutes (37°C) prior to plating. Strain specific bacterial toxicity (reduced titer) was indicated at 50 µg/plate and higher in trial one. No bacterial toxicity was noted in the repeat trial through 5000 µg/plate. There was no increase in

the mutation frequency. Positive controls were functional. Acceptable. Data are supplemental to MKH 6561. (Green and Gee, 7/26/05).

**52958-0024 214208, "Bissulfonylurea - MKH 6561, *Salmonella*/Microsome Test, Plate Incorporation and Preincubation Method", (B. Herbold, Bayer AG, Department of Toxicology, Wuppertal, Germany, Bayer AG Report No. 28896, Bayer AG Study No. T 7059968, Test No. 109255, 24 June 1999). Triplicate cultures of *Salmonella typhimurium* LT2 strains TA98, TA100, TA102, TA1535, and TA1537 were exposed to MKH 6561 (97.2% propoxycarbazone-sodium), in the presence and absence of S9, at 0 (DMSO), 16, 50, 158, 500, 1581, and 5000 µg/plate for 48 hours at 37°C. Two trials were performed. The direct plate incorporation method was used in the first trial. In the second trial, cultures were preincubated for 20 minutes (37°C) prior to plating. Positive controls were functional. Bacterial toxicity (reduced titer) was noted at 1581 and 5000 µg/plate in the plate incorporation assay. No increase in the mutation frequency. Acceptable. (Green and Gee, 7/22/05).

CHROMOSOME EFFECTS

**52958-0027 214211, "KTS 9061 (Metabolite of MKH 6561), *In Vitro* Chromosome Aberration Test with Chinese Hamster V79 Cells", (B. Herbold, Bayer AG, Department of Toxicology, Wuppertal, Germany, Bayer AG Report No. PH-28555, Bayer AG Study No. T5059920, Test No. 108983, 5 March 1999). Chinese hamster V79 cells were seeded in duplicate in 20 ml of medium per 75 cm^2 flask and exposed to KTS 9061 (98%) in the presence and absence of S9 at 0 (DMSO), 625, 1250, and 2500 µg/ml for 4 hours. Cells were harvested and evaluated 18 hours after the start of treatment and additionally at 30 hours for the control and 2500 µg/ml cultures. A repeat trial was performed at the same treatment levels, in the absence of S9, with 18 hour treatment and harvest times. 100 metaphases per culture were evaluated by light microscopy. The mitotic index was determined by evaluating 1000 cells per culture. Positive controls were functional. Aberrant metaphases were not increased and there was no reduction in the mitosis rate. Acceptable. These data are supplemental to MKH 6561. (Green and Gee, 7/26/05).

**52958-0042 214226, "MKH 6561, Micronucleus Test on the Mouse", (B. Herbold, Bayer AG, Department of Toxicology, Wuppertal, Germany, Bayer Report No. 23922, Study No T 1055722 + T 0055721, Test No. 107407, 4 April 1995). 5 Hsd/Win: NMRI mice per sex per group received a single intraperitoneal injection of MKH 6561 at 0 (0.5% aqueous Cremophor) and 2500 mg/kg followed by bone marrow retrieval 16, 24, and 48 hours later. Apathy, roughened fur, spasm, difficulty in breathing, and slitted eyes were recorded for treated animals through 24 hours post-dosing. There were no clinical signs thereafter. No animals died. Dose selection was based on a preliminary test. 1000 polychromatic erythrocytes were evaluated per animal using light microscopy. Normochromatic erythrocytes were also evaluated for number and micronuclei. Cyclophosphamide (positive control at 24 hours) was functional. No increase in micronucleated polychromatic erythrocytes. Acceptable. (Green and Gee, 7/22/05).

**52958-0053 214237, "MKH 6561, *In Vitro* Mammalian Chromosome Aberration Test with Chinese Hamster V79 Cells", (B. Herbold, Bayer AG, Department of Toxicology, Wuppertal, Germany, Bayer AG Report No. 24787, Bayer AG Study No. T 7054008, Test No. 108878, 6 February 1996). 1 x 10^6 Chinese hamster V79 cells were seeded in duplicate in 20 ml of medium per 75 cm^2 flask and exposed to MKH 6561 (98.8%) in the presence and absence of S9 at 0 (DMSO), 0 (untreated), 500, 2500, and 5000 µg/ml for 4 hours. Cells were harvested and evaluated 18 hours after the start of treatment and additionally at 30 hours for the 5000 µg/ml cultures. 0.2 ml Colcemid-solution (40 µg/ml water) were added to each flask 2 hours prior to harvest. Two slides per culture were prepared. 100 metaphases per culture were evaluated by light microscopy. Positive controls were functional. Aberrant metaphases were not increased by treatment with MKH 6561 and there was no indication of cytotoxicity. Acceptable. (Green and Gee, 7/22/05).

DNA DAMAGE

**52958-0047 214231, "MKH 6561, Test on Unscheduled DNA Synthesis in Rat Liver Primary Cell Cultures *In Vitro*", (S. Brendler-Schwaab, Bayer AG, Department of Toxicology, Wuppertal, Germany, Bayer AG Report No. 25197, Bayer AG Study No. T 8054009, Test No. 108879, 19 June 1996). Hepatocyte cultures (3.75×10^5) were treated in triplicate with MKH 6561 (97.8%) in the presence of ^{3}H-thymidine at 0 (DMSO), 25, 100, 250, 500, 1000, 2000, and 4000 μg/ml for 16 to 24 hours. 50 cells per slide were evaluated. Cytotoxicity was indicated at 2000 and 4000 μg/ml by trypan blue dye exclusion. Net grains per nucleus and the number of cells in repair were not increased by treatment. The positive control (2-AAP) was functional. Acceptable. (Green and Gee, 7/27/05).

METABOLISM

**52958-0061 214252, "[Phenyl-UL-^{14}C]MKH 6561: Absorption, Distribution, Excretion, and Metabolism in the Rat Including Whole Body Autoradiography", (H. Printz, Bayer AG, Agrochemical Division, Plant Protection Development Institute for Metabolism Research and Residue Analysis, Leverkusen, Germany, Bayer Study No. M 81819054, Bayer Report No. 108305, 8 January 1998). 4 male (tests 1, 2, 4, and 5) and 4 female (test 3) Wistar BOR:WISW (SPF Cpb) rats per group received a single oral gavage dose of [Phenyl-UL-^{14}C] MKH 6561 at 2 or 200 mg/kg (test 5 only). Animals in test 4 received 14 consecutive unlabelled daily doses prior to the radiolabelled one at 2 mg/kg. For whole body autoradiography, 5 males received a single radiolabelled dose by oral gavage at 2 mg/kg in test 6 and one male received a single intravenous dose (2 mg/kg) in test 7. In tests 8 and 9, six bile cannulated males per test received a single radiolabelled intraduodenal dose at 2 mg/kg. Expired CO_2 was collected for each animal for the intervals 0-4, 4-8, 8-24, 24-48, 48-72, 72-96, 96-120, 120-144, and 144-168 hours post-dosing in test 1. Urine was collected at the post-dose intervals 0-4, 4-8, 8-24, and 24-48 hours in tests 1 through 5 and additionally, in test 1, at 48-72, 72-96, 96-120, 120-144, and 144-168 hours. Feces were collected at the intervals 0-24 and 24-48 hours after dosing in tests 1 through 5, plus at 48-72, 72-96, 96-120, 120-144, and 144-168 hours in test 1. Plasma levels of radioactivity were determined at 5, 10, 20, and 40 minutes and at 1, 1.5, 2, 3, 4, 6, 8, 24, 32, and 48 hours after treatment in test 2 through 5. Sacrifice times for animals in test 1 and in tests 2 through 5 were 168 hours and 48 hours after treatment respectively. After sacrifice, in tests 2 through 5, radioactivity was determined in erythrocytes, spleen, gastrointestinal tract (GIT), liver, kidney, testis, heart, lung, skin, and carcass. For whole body autoradiography, animals were sacrificed 1, 4, 8, 24, and 48 hours post-dose in test 6, and 5 minutes post-dose in test 7. In tests 8 and 9 with bile cannulated rats, radioactivity was measured in urine and bile at 1, 2, 3, 4, 6, 8, 12, 18, and 24 hours (test 8 only) post dose and, in feces, during 24 hours. Animals were sacrificed 24 hours after treatment and radioactivity in the organs, skin, carcass, and GIT was determined. Liquid scintillation counting (LSC) was used to quantify radioactivity in samples. High performance liquid chromatography (HPLC) and mass spectrometry were used to identify MKH 6561 and metabolites. Results. Maximum radioactivity levels in plasma were reached at 20 minutes (tests 2, 3, and 4, 2 mg/kg) and at 1 hour (test 5, 200 mg/kg) post treatment. Terminal elimination half lives determined for total radioactivity in plasma ranged between 8.95 and 11.4 hours. In Test 1, means of 26.9% and 71.6% of administered radioactivity were excreted in urine and feces respectively after 168 hours. Most (26.3% in urine and 70.5% in feces) was excreted by 24 hours. In tests 2, 3, 4, and 5, mean cumulative excretion of radioactivity (% of Dose) after 48 hours in urine was 26.5%, 31.2%, 24.9%, and 20.9% respectively and 70.6%, 66.5%, 76.3%, and 82.6% in feces respectively. The majority of radioactivity was excreted by 24 hours. Absorption was slightly higher for females (resulting in higher renal excretion, test 3) compared to males after 2 mg/kg and reduced/delayed after a 200 mg/kg dose (in males, test 5). Based on renal excretion 21% to 31% of dose was absorbed. Elimination was fast with no evidence for bioaccumulation. Residues of radioactivity in organs and tissues at sacrifice (48 hours) in tests 2 through 5 were very low (0.0001% to 0.108% of administered dose). Metabolites were quantified in excreta for test 1 through 5. The main

component identified in urine (19.7% to 28.3% of dose) and feces samples (52.2% to 72.5%) was unchanged MKH 6561. Additionally, two metabolites (schematics provided) were identified in urine samples, MKH 7283 (0.1% to 0.2%) and MKH 7284 (0.9% to 1.7%). STJ 4934 was identified at 2.3% to 8.8% in feces extracts. A second metabolite, PlZ 1139 (a methyl ester of the acid resulting from hydrolysis of MKH 6561), was found in feces at 2.7% to 15.4%. It was determined that this compound had been formed artificially from the parent compound by using MeOH during sample preparation. None was detected in samples extracted without the use of MeOH. Schematics for the proposed metabolic pathway for MKH 6561 in rats after oral administration were included. In whole body autoradiography, the time course distribution of radioactivity in organs and tissues correlated well with the distribution and elimination observed in plasma, urine, and feces. Autoradiograms were included. The first bile fistulation experiment (test 8) was repeated because results were not considered reliable (high amounts of radioactivity were measured in the organs and tissues of the animals and recovery of total radioactivity amounted to 128% of administered radioactivity) compared to tests using the oral route. Results from the repeat test with bile cannulated rats (test 9) correlated well to oral route results. The major portion of administered radioactivity was excreted in the feces (87.4%).
13.3% was excreted in urine and 3.4% in bile. On average, 97% to 104% pf administered dose were recovered in excreta and organs/tissues. Acceptable. (Green and Gee, 7/26/05).

52958-0057 214248, "[Phenyl-^{14}C] MKH 6561: Occurrence of the Plant Metabolite 2-hydroxy-MKH 6561 in Excreta and Liver Extracts of the Rat", (A. Klempner, Bayer AG, Crop Protection Research Center, Institute for Metabolism Research and Residue Analysis, Leverkusen, Germany, Bayer Study No. M71819071, Bayer Report No. 109200, 17 June 1999). 4 male Wistar BOR:WISW (SPF Cpb) rats per group received a single oral gavage dose of [Phenyl-UL-^{14}C] MKH 6561at 2 mg/kg followed by sacrifice 20 and 90 minutes later. At sacrifice (20 minutes (Test 1) and 90 minutes (Test 2) after dosing), collected blood was separated into plasma and erythrocytes by centrifugation. Radioactivity was quantified in plasma, urine, feces, liver, skin, carcass, and the gastrointestinal tract (GIT) by liquid scintillation counting (LSC). Livers were extracted. HPLC fractionation of liver extracts followed by further HPLC and TLC analysis of the HPLC fractions were used to identify 2-hydroxy-MKH 6561. Additionally, urine pools and feces extracts from the Rat Metabolism Study (Study No. M 81819054, record 214252, volume 52958-0061) were analyzed for 2-hydroxy-MKH 6561. 2-hydroxy-MKH 6561 was identified in dosing solutions at 0.02% to 0.04%. Radioactivity in liver fractions represented 2.8 % and 5.7% of the administered dose in tests 1 and 2 respectively. 2-hydroxy-MKH 6561 was identified at < 0.01% of administered dose in test 2. It was not detected in test 1. Excretion of radioactivity in urine and feces was insignificant. During 20 minutes (test 1) urine excretion accounted for < 0.01% of administered radioactivity. 90 minutes after dosing (test 2), 2.3% of radioactivity was excreted in urine and < 0.01% in feces. 2-hydroxy-MKH 6561 was not detected in feces extracts generated from the Rat Metabolism Study (Study No. M 81819054, record 214252, volume 52958-0061). In urine pools from that study (collected 0-24 hours post-dosing), excreted radioactivity represented 20.7% to 29.9% of administered dose and 2-hydroxy-MKH 6561 was found (HPLC) at 0.04% to 0.10%. The conclusion was that 2-hydroxy MKH 6561 in the administered dose was excreted in the urine and was not a metabolite in liver of rats. Acceptable as a supplementary study. (Green and Gee, 7/27/05).

**52958-0063 214254, "[Triazolinon-3-^{14}C]MKH 6561: Absorption, Distribution, Excretion and Metabolism in the Rat", (H. Printz, Bayer AG, Agrochemicals Division, Plant Protection Development Institute for Metabolism Research and Residue Analysis, Leverkusen, Germany, Bayer Study No. M 11819057, Bayer Report No. 108304, 1 December 1997). 4 male Wistar BOR: WISW (SPF Cpb) rats per group received a single oral gavage dose of [triazolinon-3-^{14}C] MKH 6561 at 2 mg/kg. 3 Tests were performed. Radioactivity in expired air, urine, feces, and the carcass was determined for Test 1 animals. Radioactivity in urine, feces, plasma, and organs was evaluated in Tests 2 and 3 (repeat of test 2). Urine was collected at the intervals 0-4, 4-8, 8-24, and 24-48 hours post-dosing. Feces and expired air (Test 1 only) were collected at 0-8, 8-24, and 24-48 hours after dosing. At sacrifice (48 hours after dosing), animals were exsanguinated

and collected blood was separated (plasma and erythrocytes) by centrifugation. Radioactivity was quantified in urine, feces, plasma, erythrocytes, spleen, liver, kidney, testis, skeletal muscle, bone, heart, lung, skin, gastrointestinal tract (GIT), and carcass by liquid scintillation counting (LSC). The majority of radioactivity was excreted in the feces and urine during 24 hours post-dosing. 70.7%, 65.1%, and 62.4% of administered radioactivity was recovered in feces and 23.3%, 21.8%, and 24.0% in urine during 24 hours in Tests 1, 2, and 3 respectively. Expired air (Test 1) accounted for 0.153% of radioactivity during 48 hours after treatment. Concentrations of total radioactivity in organs and tissues at sacrifice (48 hours post-dosing) were very low (< 0.1%). Peak plasma concentrations of radioactivity were recorded 0.32 hours after treatment. Terminal elimination half-lives for total radioactivity in plasma ranged from 12 to 13 hours indicating a low potential for bioaccumulation in organs and tissues. Metabolites were identified by high performance liquid chromatography (HPLC). In feces, 58% to 66% of radioactivity excreted over 24 hours represented MKH 6561. An additional metabolite was identified as MKH 7017 at 3%. During the 24-48 hour interval, MKH 6561 and MKH 7017 accounted for 1.6% and 0.1% of administered dose respectively. The majority of radioactivity excreted in urine over 24 hours (22% to 23%) represented MKH 6561. Up to 4 metabolites (all more polar than MKH 6561) were observed (but not identified) in urine samples. Each accounted for less than 1% of administered radioactivity and combined they accounted for less than 1.5% of the total. A diagram of the proposed metabolic pathway for MKH 6561 was included. Acceptable. (Green and Gee, 7/27/05).

SUBCHRONIC STUDIES

Rat 4-Week Dietary Toxicity Study

0020; 214204; “MKH 6561 Study for Subacute Toxicity in Rats (Feeding Study)” (Krötlinger, F., Bayer AG, Department of Toxicology, Wuppertal, Germany, Bayer AG Report No. 25104, Bayer AG Study No. T2055651, 05/15/96). MKH 6561 (Batch No. NLL 5551-22.1, purity = 97.8%) was admixed to the diet (containing 1% peanut oil) and fed to 5 Wistar (Hsd Win:WU) rats per sex per dose at dose levels of 0 (untreated diet), 800, 4000, 10000, or 20000 ppm (0, 79.6, 391.1, 1074.4, and 2146.2 mg/kg/day, respectively for males and 0, 79.1, 399.0, 985.4, and 2305.3 mg/kg/day, respectively for females) for 28 days. No mortalities occurred. No clinical signs were observed. No treatment-related body weight effects were observed. Hematology and serum chemistry revealed no treatment-related effects. Organ weight data revealed no treatment-related effects. Macroscopic and microscopic examinations revealed no abnormalities. NOEL (M) = 2146.2 mg/kg/day (20000 ppm) and NOEL (F) = 2305.3 mg/kg/day (20000 ppm) no effects at the highest dose tested (note: the presented immunotoxicity investigations were reviewed but the results of these investigations were not included when establishing a NOEL due to difficulty in interpretation). **Supplemental study** (only 5 animals per sex per dose level were used and the animals were treated for only 28 days). (Corlett, 05/26/05)

Rat Subchronic Dietary Toxicity Study

**52958-0050 214234, “MKH 6561, Study for Subchronic Oral Toxicity in Rats (Feeding Study for 14 Weeks with a 4-Week Recovery Period)”, (W. Bomann, P. Andrews, and B. Schilde, Bayer AG, Department of Toxicology, Wuppertal, Germany, Bayer AG Report No. 25597, Bayer Report No. 108881, 26 September 1996). 10 Wistar rats per sex per group received MKH 6561(97.80% propoxycarbazone) in the diet at 0 (standard diet), 250, 1000, 4000, and 20000 ppm for 14 weeks. Ten additional control and high dose rats per sex per group received test diets for 14 weeks followed by a 4-week recovery phase where they received untreated standard diet. The mean dietary intake of MKH 6561 was 17.4, 73.0, 286.4, and 1507.5 mg/kg/day for males and 21.6, 81.6, 350.6, and 1769.9 mg/kg/day for females at 250, 1000, 4000, and 20000 ppm respectively. Water intake was increased (statistically significant) at 20000 ppm for both sexes during treatment and was comparable to controls during the recovery period. Glucose and triglyceride levels were significantly reduced in high dose females. Clinical signs, mortality, bodyweight, food consumption, ophthalmology, hematology, urinalysis, necropsy, and organ weights were not affected by treatment. Microscopy indicated increased incidence of vacuoles in the squamous epithelium of the forestomach along with minimal epithelial hyperplasia and subepithelial inflammatory infiltration for main group animals that received 20000 ppm. These

effects reversed during the 4-week recovery period. NOEL = 4000 ppm (increased water intake, reduced blood triglycerides and glucose in females, and forestomach epithelium irritation). No adverse effects. Acceptable. (Green and Gee, 7/28/05).

Rat Subchronic Neurotoxicity Study

0049; 214233; "MKH 6561 Subchronic Neurotoxicity Screening Study in Wistar Rats (Thirteen-Week Administration in the Diet)" (Dreist, M. and Popp, A., Bayer AG, Department of Toxicology, Wuppertal, Germany, Bayer AG Report No. 27779, Bayer AG Study No. T 9061615, 08/13/98). 827. Technical Grade MKH 6561 (Batch No. 05649/0004, purity = 98.0%) was admixed to the feed (containing 1% corn oil by weight) and fed to 12 Wistar (Hsd Cpb:WU) rats per sex per dose at dose levels of 0 (untreated diet), 1000, 4000, or 20000 ppm (0, 64, 252, and 1321 mg/kg/day, respectively for males and 0, 79, 312, and 1651 mg/kg/day, respectively for females) for 13 weeks. No mortalities occurred. No treatment-related clinical signs were observed. No treatment-related effects on body weight was observed. FOB, motor activity, and locomotor activity assessments revealed no treatment-related effects. Macroscopic and neuropathological examinations revealed no treatment-related abnormalities. **No adverse effects.** NOEL (M)= 1321 mg/kg/day (20000 ppm) and NOEL (F) = 1651 mg/kg/day (20000 ppm) based on no treatment-related effects at the highest dose tested. **Acceptable.** (Corlett and Leung, 06/10/05)

Rat 4-Week Repeated Dosing Dermal Toxicity Study

0030; 214214; "MHK 6561, Study for Subacute Dermal Toxicity in Rats (Four-Week Treatment Period)" (Krötlinger, F. and Schilde, B., Bayer AG, Department of Toxicology, Wuppertal, Germany, Bayer AG Study No. T1060960, Bayer AG Report No. 26429, 07/01/97). 822. MKH 6561 (Mixed batch no. 05649/0004, purity = 97.6%) was moistened with tap water and applied to the shaved dorsal skin of 5 HsdCpb:WU rats per sex per dose at dose levels of 0 (tap water only) or 1000 mg/kg/day for 6 hours per day 5 days per week for 4 weeks (7 days per week during the 4th week). No mortalities occurred. No clinical signs were observed. No skin irritation was observed at the treatment sites. No effect on body weight was observed. Hematological and clinical chemistry investigations revealed no treatment-related effects. Statistically significant increases in mean absolute and mean relative lung weights in males at 1000 mg/kg/day was observed. Microscopic examination revealed an increase in frequency and severity of focal inflammation in the lungs of males at 1000 mg/kg/day. **No adverse effects.** NOEL (M, systemic) < 1000 mg/kg/day (based on increased mean absolute and mean relative lung weights and an increase in frequency and severity of focal inflammation in the lungs) and NOEL (F, systemic) = 1000 mg/kg/day based on no effects at the highest dose tested. NOEL (M/F, skin) = 1000 mg/kg/day based on no treatment-related effects at the highest dose tested. **Acceptable.** (Corlett, 06/16/05)

Mouse 5-week Dietary Toxicity Study

0019; 214203; "MKH 6561 Subacute Toxicity Study in B6C33F1-Mice (Administration in the Feed Over 5 Weeks)" (Schadt, L., Bayer AG, Department of Toxicology, Wuppertal, Germany, Bayer AG Report No. 25341, Bayer AG Study No. T4055699, 08/12/96). MKH 6561 (Batch No. NLL 5551-22.1, purity = 97.8%) was admixed to the diet (containing 1% peanut oil) and fed to 5 $B6C3F_1$ mice per sex per dose at dose levels of 0 (untreated diet), 100, 1000, or 10000 ppm (0, 41.7, 393.8, and 5579.4 mg/kg/day, respectively for males and 0, 63.9, 546.6, and 5989.1 mg/kg/day, respectively for females) for 5 weeks. No mortalities occurred. No clinical signs were observed. A slight decrease (not statistically significant) in body weight gain was observed in both sexes at 10000 ppm. Hematology and serum chemistry revealed no treatment-related effects. Organ weight data revealed no treatment-related effects. Macroscopic examinations revealed no abnormalities. NOEL (M) = 5579.4 mg/kg/day (10000 ppm) and NOEL (F) = 5989.1 mg/kg/day (10000 ppm) based on no treatment-related effects at the highest dose tested. **Supplemental study** (only 5 animals per sex per dose level were used, no ophthalmological examinations were conducted, the animals were treated for only 5 weeks, and no histopathological examinations were performed). (Corlett, 05/20/05)

Mouse Subchronic Dietary Toxicity Study

52958-0021 214205, "MKH 6561, Subchronic Toxicity Study in B6C3F1-Mice (Administration in the Feed Over 14 Weeks)", (L. Schadt and V. Geiss, Bayer AG, Department of Toxicology, Wuppertal, Germany, Bayer AG Report No. 25796, Bayer AG Study No. T1055704, Test No. 108877, 6 January 1997). 10 $B6C3F_1$ mice per sex per group received MKH 6561 (97.8% propoxycarbazone-sodium) in the diet at 0 (standard diet), 625, 2500, and 10000 ppm for 14 weeks. The average MKH 6561 intake during the treatment period was 205.1, 860.2, and 3926.2 mg/kg/day for males and 306.9, 1158.8, and 5108.9 mg/kg/day for females at 625, 2500, and 10000 ppm respectively. Group mean food consumption was slightly increased for males at 10000 ppm. Male and female bodyweights were significantly lower than controls at 2500 and 10000 ppm. Cholesterol was significantly decreased for 10000 ppm males relative to concurrent controls, but within the historical control range. Other serum chemistry parameters were comparable to controls. The leucocyte count was significantly lower than controls in treated males and in high dose females. No treatment-related changes were recorded for the other hematology parameters. Group mean relative liver weights were reduced (ns) in males at 2500 and 10000 ppm relative to controls. Gross pathology and histopathology were unremarkable. NOEL = 625 ppm based on reduced bodyweights. No adverse effects were indicated. Unacceptable, not upgradeable (incomplete histopathology, no ophthalmology). (Green and Gee, 7/28/05)

**52958-0051 214235, "MKH 6561, Plaque-Forming -Cell Assay in Rats (Feeding Study Over About 4 Weeks)", (F. Krotlinger and H.W. Vohr, Bayer AG, Department of Toxicology, Wuppertal, Germany, Bayer AG Report No. PH 29238, Bayer AG Study No. T9068906, Test No. 109408, 26 October 1999). 8 male Wistar HsdCpb:WU rats per group received MKH 6561 (98.3%) in the diet at 0 (standard diet), 4000, 10000, and 20000 ppm for 4 weeks. Animals were immunized by intravenous injection of the T-cell dependent antigen, sheep erythrocytes (SRBC) (100 μl of a solution of 5×10^8 per ml), 5 days prior to the end of treatment in preparation for the plaque-forming cell (PFC) response assay. Average MKH 6561 intake during the treatment period was 401, 986, and 2144 mg/kg/day at 4000, 10000, and 20000 ppm respectively. Water intake was significantly increased at 20000 ppm. Discolored feces were noted for one and 6 animals at 4000 and 20000 ppm respectively during the treatment period. At the end of treatment, animals were sacrificed, spleens were removed and crushed through a metal sieve resulting in single cell suspensions. Four aliquots of the suspensions (at two times 100 μl and 10 μl) were used for detection of SRBC specific B cell activation. SRBC specific IgM plaques were determined for each animal in duplicate on glass slides and the amount of plaque forming cells (PFC) per 10^6 spleen cells was calculated. No treatment-related effect on mean cell counts ($x10^6$) of the spleen. Group mean counts of plaque forming cells per 10^6 spleen cells for treated groups were in a range comparable to control values. Immunotoxicity NOEL = 20000 ppm. Clinical NOEL = 10000 ppm based on increased water intake. Acceptable. This study is not a data requirement at this time. It is supplemental data to MKH 6561. (Green and Gee, 7/27/05).